the plants survived, and bore 72 leaves, blossoming about January 1. Considerable seed was saved from both the terminal and lateral inflorescence of this plant.

In 1913 about 5,000 plants were grown from this seed. These plants were true to the new type in all external characters, and differed from the normal Cuban in having a somewhat lighter green shade to the leaves, in an absence of basal suckers (lateral branches), and in a practically indeterminate growth, whereas the normal Cuban variety produces a terminal inflorescence after producing from 16 to 25 leaves on the main stem. Twenty plants were brought to our greenhouse in New Haven; all but eight, however, were injured during transportation. The eight uninjured plants commenced to blossom about the first of November, the range of leaf counts per plant being from 62 to 80, with the greater number around 70. These data show that this tobacco mutant is breeding true, and unless it behaves in a different manner from other mutants, it should breed true in succeeding generations.

The cured leaves are very promising, resembling the normal variety. There is every reason to believe that this new type will prove of commercial value, as the yield per acre is at least fifty per cent. greater than the normal type. It has been named the Stewart Cuban.

The normal Cuban seed which was saved in 1910 was again used for planting in 1913, and over 200 acres, or two and one half million plants, were grown. Although search was made at the Windsor Tobacco Growers' Corporation, which grows over 100 acres, no mutating plants were discovered. Two mutants were found at other plantations where the 1910 Windsor Corporation seed was used, which presented the same habit of producing a large leaf number. Thus, five similar mutants from the same seed have been discovered, though it can not be stated that they did not all come from a single normal plant. The frequency of the appearance of this mutation is at the rate of about one plant in a million.

This mutation must have taken place after fertilization, *i. e.*, after the union of the male and female reproductive cells. If the mutation had taken place in either the male or female cell before fertilization, the mutant would have been a first generation hybrid, and would have given a variable progeny the following season.

Mutations of high leaf number have been observed in tobacco previous to this time. Several years ago a variant with a large leaf number was found in the outdoor Havana type at the farm of Mr. Alsop in Avon, and in 1912 a Havana plant which bore 72 leaves was found at the Olds Brothers' Plantation in Bloomfield. Six similar mutations were found at a Windsor farm, and one at another farm in Bloomfield this last season. It is of interest to know that these mutations occurred in a variety, the Connecticut Havana, which has been grown in Connecticut for a period of over fifty years, and which is very uniform in habit. That it has been observed in different sections and by different growers shows that the same mutation must have taken place several times.

A similar type bearing a large leaf number appeared in Maryland several years ago, and is grown commercially under the name of Maryland Mammoth. The Maryland type was, however, the result of a cross between two Maryland tobacco varieties. The mutations which have occurred in Connecticut can hardly be explained on the basis of the results of a cross.

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REAL AND APPARENT NITRIFYING POWERS

IN making bacteriological studies of soils one of the leading factors determined is the ability of the bacteria present to convert various forms of nitrogen into the nitrate condition. Comparison of this ability, as existing in different soils, is usually made under definite, more or less standard conditions. This factor is spoken of as the "nitrifying power" or "nitrifying efficiency."

Of recent years the tendency has been to employ the soil to be tested, or a standard soil, as the medium in which the organisms

shall work. An additional supply of nitrogen is usually furnished, either in the form of ammonium-sulphate, or some highly nitrogenous, easily decomposed, organic substance, such as cottonseed meal or tankage. Where soil has been employed as the medium, one of three methods of procedure has usually been followed. One method is to add to a definite weight of soil a given amount of nitrogen, incubate and then determine the quantity of nitrate nitrogen present. The amount found, taking no account of the nitrate nitrogen originally present, is regarded as indicating the comparative ability of different soil samples to form nitrate nitrogen from other forms of nitrogen; in other words, its nitrifying power—ability—efficiency. A second method is to make a determination of the amount of nitrate nitrogen present in a corresponding sample at the beginning of the experiment; otherwise proceed as above. The difference between the amount originally present and that found at the final analysis is regarded as the correct factor. By a third method duplicate samples are taken and treated exactly alike with the exception that only one receives an additional supply of nitrogen. After incubation both are analyzed and the difference in the nitrate nitrogen content is regarded as the correct factor for comparison.

Recently, in determining what Stevens and Withers have termed the nitrifying inoculating power of a series of plots that are under study, it became necessary to use a soil relatively high in nitrate nitrogen. Cottonseed meal was employed as the source of nitrogen. In a large number of cases, securing the comparative factor by either of the last two mentioned methods, it was found to be a minus quantity. That is, with the conditions optimum (so far as is known) for nitrification, the amount of nitrate nitrogen found after incubating was much less than originally present in the soil. If ammonium sulphate were substituted for cottonseed meal, as the source of nitrogen, such a condition was never noted. This led to a series of investigations to determine the cause of the minus factors that were secured. There were evidently losses of large quantities of nitrate nitrogen. Was this due to actual losses of total nitrogen or was the nitrate nitrogen merely transformed? To what extent does it occur? What are the conditions influencing the same? These are some of the questions an effort is being made to answer. Certain grave difficulties have thus far prevented a complete solution of the problems involved. However, it is believed that in the near future sufficient data, together with that upon which the statements herein presented are made, will be secured to completely settle the question.

One very interesting fact has been brought to light. This is that at least two of the methods now employed in bacteriological laboratories for determining the relative nitrifying power of soils may give us absolutely no indication of true values. The writer has abundant evidence to show that this is true in the presence of appreciable quantities of nitrate nitrogen when easily decomposed organic substances are employed as the source of nitrogen. Oft-repeated observations show that where from five to twenty-five mg. of NO_3 per 100 gr. soil is present and cottonseed meal added, at the end of seven days absolutely no trace of NO_3 can be found. If examined three weeks later an abundance will be present. According to the last two methods mentioned above, the five to twenty-five mg. NO_3 would be subtracted from that found at the final analysis and the difference taken as the factor indicating the amount formed during incubation. In reality, the amount present reached zero at one time, hence all found at the final analysis must have been formed during incubation, and represents more nearly the correct factor.

In a series of experiments designed for the purpose of determining how much NO_3 would disappear it was found that in five days in the presence of .95 gr. cottonseed meal, as high as thirty-five mg. NO_3 per 100 gr. soil had been lost. With 1.9 gr. cottonseed meal the quantity ran up to fifty-five mg. With higher quantities of cottonseed meal still larger amounts of NO_3 were lost. With

small quantities of cottonseed meal the nitrate nitrogen may begin to accumulate again after ten days' incubation. With larger quantities the time is longer. This fact has probably caused many to overlook the first disappearance.

Just what becomes of the nitrate nitrogen, under such conditions, has not been determined. There are two possibilities. It may be liberated in the elementary form through the process of denitrification. This seems improbable since the soil in the writer's experiments has never been much over an inch in depth and never more than two thirds saturated, hence aeration was good. Another possibility is that it may be assimilated. There is always a very copious growth of soil fungi of various forms when cottonseed meal is applied. In fact, so abundant are the mycelial threads that they bind the soil together in a mass which is rather difficult to disintegrate by the ordinary shaking method. It is highly probable that at least a portion of the nitrate nitrogen is assimilated by this growth. It would seem that it would be very easy to determine the above question, but the volatilization of ammonia where large amounts are being formed, as is always the case in the presence of cottonseed meal, makes it difficult.

It is evident, from what has just been said, that deducting the nitrate nitrogen originally present or that in an incubated check, will not give us correct results under the conditions mentioned above. The more nitrate nitrogen initially present, the less reliable will be our results. These two methods must then be abandoned. Simply taking as the correct factor the amount found at the final analysis will probably approach nearer the truth than any other method now in practise. However, we have no assurance that this gives us an accurate idea of the relative amount formed in different soils. There is absolutely no way of determining the actual amount formed that immediately disappears. We only know that in this method the actual amount present at one time (unless very large amounts were initially present) was zero, and that all formed at any future date must have been formed during the course of the experiment.

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ON THE APPARENT ABSENCE OF APOGAMY IN ŒNOTHERA

IN a previous note in SCIENCE[1] I described certain experiments which suggested that *Œnothera* was occasionally apogamous. Three imperfect seeds were obtained from a castrated flower of *O.* mut. *lata,* suggesting the possibility that a small percentage of the seeds might develop apogamously. Last year (1912) the experiments were carried out more extensively, however, and the results were wholly negative, showing that if apogamy occurs in *O.* mut. *lata* it must be very rare indeed.

Six *lata* plants were experimented upon, whose history was as follows: one was a mutant appearing in a culture of a race of *O. Lamarckiana* from the Kolosvar Botanical Garden; two were derived from *lata* self-pollinated in the cultures of de Vries; two were mutants occurring in a *rubinervis*-like race obtained from Heribert-Nilsson in Sweden; and one was *O. biennis* mut. *lata* appearing in a race of *O. biennis* from the Madrid Botanical Garden. On these plants over 20 flowers were castrated and covered with bags during the height of the blooming season, from July 22 to August 13, 1912. In addition, a whole branch of one pollen-sterile *lata* plant, containing 20 flowers, was covered with a large bag. No growth of the capsules took place in any case, and not a single seed could be found in any of the capsules. It would appear, therefore, that under these circumstances at least, apogamous development practically never occurs in *lata,* for the number of ovules in the capsules observed must have numbered several thousands. The plants were, moreover, all well nourished.

Similar experiments made with eight flowers belonging to four plants of *O.* mut. *gigas* also

[1] Gates, R. R., "Apogamy in *Œnothera*," SCIENCE, N. S. 30: 691–694, 1909.